purification and stabilization procedures were developed for each of the LASV proteins individually, as each polypeptide required specific conditions for optimal purification and solubility in aqueous solutions. Optimal expression, purification, and solubilization formats were subsequently chosen for each LASV protein, and the processes were standardized. Specific examples are outlined in the body of this application.

SUMMARY OF INVENTION

[0014] The present invention discloses compositions comprising soluble and membrane-anchored forms of Lassa virus (LASV) glycoprotein 1 (GP1), glycoprotein 2 (GP2), the glycoprotein precursor (GPC), and the nucleocapsid protein (NP). Another embodiment of the present invention is drawn to proteins that consist of soluble and membrane-anchored forms of Lassa virus (LASV) glycoprotein 1 (GP 1), glycoprotein 2 (GP2), the glycoprotein precursor (GPC), and the nucleocapsid protein (NP). This invention also relates to diagnostic and preventative methods using the novel forms of the LASV subunit proteins. Preventative methods include preparation of vaccines, as well as factors (e.g. small molecules, peptides) that inhibit LASV infectivity. Further, the invention relates to diagnostic and therapeutic antibodies including neutralizing antibodies for the prevention and treatment of infection by LASV and other arenaviruses. The present invention also discloses and provides new tools and methods for the design, production, and use of soluble and membrane-anchored forms of LASV GP1, GP2, NP and GPC including expression in engineered bacterial- and mammalian-based systems.

[0015] One embodiment of the invention relates to polynucleotides and polypeptides or fragments thereof encoding soluble forms of LASV GP1. The polynucleotide sequences may encode polypeptides that comprise or consist of soluble forms of LASV GP1 or fragments thereof.

[0016] Another embodiment of the invention relates to polynucleotides and polypeptides or fragments thereof encoding soluble forms of LASV GP2. The polynucleotide sequences may encode polypeptides that comprise or consist of soluble forms of LASV GP2 or fragments thereof.

[0017] Another embodiment of the invention relates to polynucleotides and polypeptides or fragments thereof encoding membrane-anchored forms of LASV GPC. The polynucleotide sequences may encode polypeptides that comprise or consist of membrane-anchored forms of LASV GPC or fragments thereof.

[0018] Another embodiment of the invention relates to polynucleotides and polypeptides or fragments thereof encoding a form of LASV NP. The polynucleotide sequences may encode polypeptides that comprise or consist of LASV NP or fragments thereof.

[0019] Another embodiment of the invention relates to methods of producing forms of LASV GP1, GP2, GPC, and NP.

[0020] Another embodiment of the invention relates to expression vectors comprising polynucleotides encoding forms of LASV GP1, GP2, GPC, and NP.

[0021] Another embodiment of the invention relates to fusion proteins comprising a polypeptide of the invention and one or more polypeptides that enhance the stability of a polypeptide of the invention and/or assist in the purification of a polypeptide of the invention.

[0022] An embodiment of the invention relates to antibodies or fragments thereof, such as neutralizing antibodies, specific for one or more polypeptides of the invention and diagnostic and/or therapeutic application of such antibodies.

[0023] Another embodiment of the invention relates to diagnostics comprising the polypeptides of the invention and/or antibodies or fragments thereof including labeled antibodies or fragments thereof of the invention.

[0024] Another embodiment of the invention relates to a subunit vaccine comprising the polynucleotides or polypeptides of the invention.

[0025] Another embodiment of the invention is directed to kits comprising the polynucleotides, polypeptides, and/or antibodies of the invention.

[0026] Other embodiments and advantages of the invention are set forth in part in the description, which follows, and in part, may be obvious from this description, or may be learned from the practice of the invention.

DESCRIPTION OF THE TABLES

[0027] Table 1 describes the oligonucleotide primers used for amplification of LASV genes for expression in *E. coli*.

[0028] Table 2 describes the oligonucleotide primers used for amplification of LASV genes for expression in mammalian cells.

[0029] Table 3 is a summary of vectors and respective *E. coli* strains used to express recombinant LASV genes.

[0030] Table 4 is a summary of vectors and respective mammalian cell lines used to express recombinant LASV genes.

[0031] Table 5 summarizes studies for invention production phase 1 as described in Example 10.

[0032] Table 6 summarizes studies for invention production phase 2 as described in Example 10.

[0033] Table 7 presents data showing that the recombinant IgM capture ELISA is a much faster assay (approximately 1.5 hours) than the traditional IgM capture assay, which takes over 6 hours (refer to Example 10).

[0034] Table 8 presents comparisons of recombinant LASV ELISA with traditional ELISA and PCR detection using a serological panel from the Kenema Government Hospital Lassa Ward (refer to Example 10).

[0035] Table 9 shows IgM and IgG reactivity to recombinant LASV proteins in a cohort of follow-up patients from the Lassa Ward of Kenema Government Hospital and their household contacts (refer to Example 10).

DESCRIPTION OF THE FIGURES

[0036] FIG. **1** depicts the phylogenetic relationships among the members of the family Arenaviridae. Partial NP gene nucleotide sequences were aligned and analyzed by maximum parsimony (redrawn from Bowen, Peters, and Nichol 1996. See also Bowen et al., 2000).

[0037] FIG. **2** depicts the cloning strategy for expression of LASV proteins (A) GP1, GP2, and NP in *E. coli* using pMAL vectors and (B) GPC, GP1, and GP2 in mammalian cells using the human cytomegalovirus (CMV) promoter-driven eukaryotic vectors. (A) To generate MBP-LASV gene fusions for *E. coli* expression, PCR-amplified LASV gene sequences were restricted and cloned in-frame at the 3' end of the malE gene, beyond the cleavage site for Factor Xa (IQGR). The LASV GP1 gene sequence comprised amino acids (a.a.) 59-259 in the native GPC, spanning the first a.a. beyond the known signal peptidase (SPase) cleavage site at position 58 to the junction between GP1 and GP2 domains,